

Full Length Research Paper

Taxonomical study of the genus *Amanita* from Western Burkina Faso

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Received 2 September, 2019; Accepted 17 October, 2019.

Burkina Faso has abundant wild mushrooms, but very few inventories have been carried out to record these species. The shortage of taxonomic studies in Burkina Faso on wild mushroom species limits their knowledge. Three species of the genus *Amanita* from Burkina Faso are reported and fully described in this paper. They are *Amanita pulverulenta* Beeli, *Amanita citrina* (Schaeffer) Persoon and *Amanita odorata* Beeli. All the three species were collected in the classified forest of Niangoloko, previously in the mosaic gallery forests dominated by *Berlinia grandiflora* (Vahl) Hutch. & Dalziel and *Isoberlinia doka* Craib & Staph (Caesalpiniaceae) and are assumed to be ectomycorrhizal.

Key words: Fungi, gallery forest, Niangoloko, Burkina Faso.

INTRODUCTION

This article discusses the genus *Amanita* Pers. as part of the in-depth study on macromycetes by many mycologists in Burkina Faso. No scientific study has yet been carried out taxonomically by a mycologist on this genus in this country and many species will disappear without being known to the scientific world. There is therefore a need to make them known through inventories. The history of work on the diversity of macromycetes in Burkina Faso can be traced to the research carried out by Heim (1936) on "Overview of Madagascar mycological flora III: Three giant boletus from Africa and Madagascar". Currently, the main focus

is to collect additional data on Burkina Faso fungi through Burkina Faso's national fungal biodiversity inventory, and to date, work carried out as part of a regional project coordinated by the University of Ouagadougou, has documented an extremely diverse macrofungal community (Sanon et al., 2017). The genus *Amanita* is a group of fungi cosmopolitan in distribution and especially well represented in tropical Africa, but most previous taxonomic studies were largely based solely on morphology (Tang et al., 2015).

Studies on *Amanita* have focused much attention on toxin-producing species (Hawkeswood, 2006; Brosseau,

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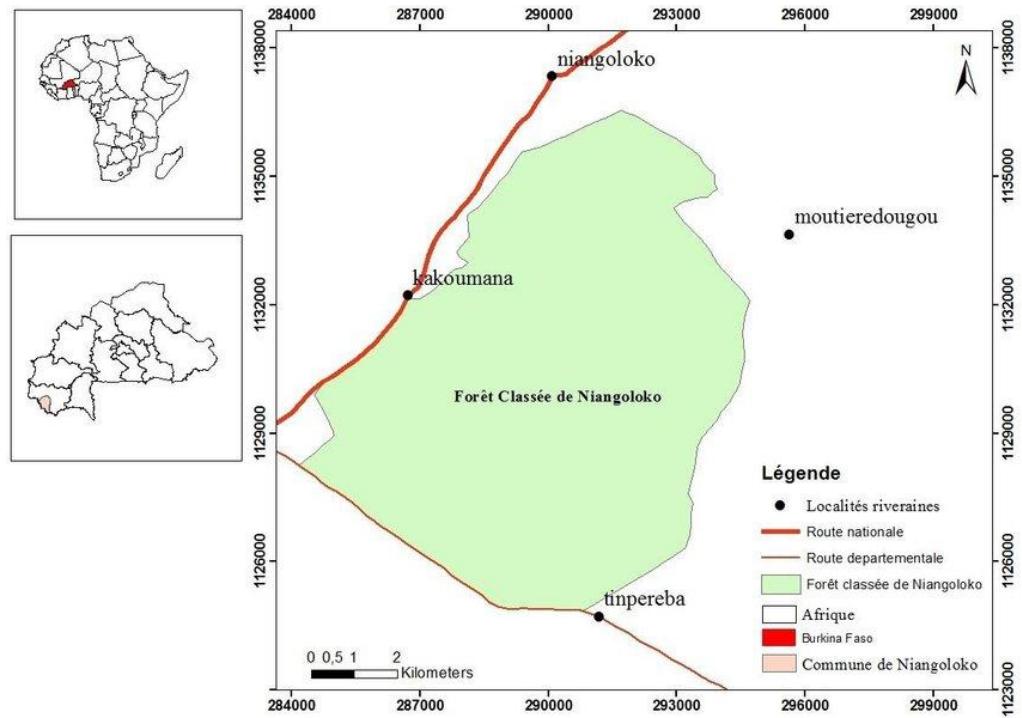


Figure 1. Geographical location of the Niangoloko forest.
Source: Ouoba et al. (2019).

1982; Cai et al., 2014; Zhang et al., 2010, 2015). There are about 58 endemic taxa in sub-Saharan Africa (Tulloss and Possiel, 2017). Indeed, the genus *Amanita* has been the subject of advanced documentation through many studies carried out in Africa: Beeli (1931, 1935), Gilbert (1940, 1941) and Bas (1969) in Congo, Bouriquet (1941, 1943) in Madagascar, Pegler and Pearce (1980) and Pegler and Shah-Smith (1997) in Zambia, Bas (1982) in Malawi, Reid and Eicker (1991) and Eicker et al. (1993) in South Africa, Häkkinen et al. (1994) in Tanzania. Up to now, our survey on Amanitaceae in Burkina Faso shows that there are at least 20 species of *Amanita* in the country; among them, a number will certainly be new to science and Burkina Faso. To date, 1000 species have been described worldwide in the Amanitaceae family (Yang et al., 2018), including about 50 species in tropical Africa. The genus *Amanita* (Amanitaceae) is described as an ectomycorrhizal genus establishing a symbiotic association with partner plants (Onguene and Kuyper, 2012). Most species in the genus are considered to be ectomycorrhizal (EcM) and they are distributed in forests and heaths including Betulaceae, Dipterocarpaceae, Fabaceae, Myrtaceae, Pinaceae, and Salicaceae. They play a critical role in forest ecosystems worldwide (Zhang et al., 2004) and thus contribute to the improvement of water and mineral nutrition of partner plants (Onguene and Kuyper, 2012).

In temperate regions, the genus has a very bad reputation due to powerful and deadly toxins present in a number of species. *Amanita* Pers. is an important

genus of mushrooms that includes several species that are widely recognized as the most toxic mushrooms in the world (Thongbai et al., 2016). However, few species are consumed. In tropical Africa, several species of edible amanites, including the very large *Amanita loosii*, appear to be widespread in open African forests (De Kesel et al., 2017).

Despite these contributions, the amanites of Burkina Faso are still very poorly regarded; they are non-existent compared to those of some other regions of the world. Further studies seem necessary to know and document the fungal diversity of Burkina Faso's natural ecosystems through inventories. Our work aims to do a taxonomic study of amanite species in Burkina Faso.

MATERIALS AND METHODS

Study area

Located in the extreme southwest of Burkina Faso, the city of Niangoloko, bordering the nature reserve, is 18 km from the border with Côte d'Ivoire. Since 1936, the Niangoloko classified forest has been part of the State's classified forest estate. It is located in southwestern Burkina Faso (Figure 1) and covers an area of 7295.83 ha between 4° 50' and 4° 58' W and between 10° 10' and 10° 17' N latitude (Ouoba et al., 2019). The natural reserve extends over an area of 7300 ha. The climate of this region, the rainiest of the country, is of Sudanian type (Guinko, 1984). The average annual rainfall is 1115 mm. The average temperature is 27.35°C with minimum of 18.3°C in December and maximum of 36.6°C in



Figure 2. Basidiomes of *Amanita citrina* show the annulus and the scales on the cap.

March (Ouoba et al., 2019). The vegetation and flora of the natural reserve of Niangoloko have been the subject of an in-depth study (Ouoba, 2006). The characteristic vegetation is a mosaic of gallery forests, shrubby savannas, wooded savannas and wooded savannas. All around the reserve, towns and villages of the Goin (farmers) and Peulh (herders) ethnic groups have settled. The main authorized activities within the forest are fruit picking, honey harvesting, medicinal plant harvesting and dead wood collection.

Collection, macroscopy and microscopy

The methodology used is similar to that of Tulloss (2008). The samples were collected in the Cascades region in July and August 2018, a timeframe during which rainy season is well established in the area. The samples were carefully collected from the ground with a pocket knife in order to obtain a complete carpophore, often necessary for the determination of a collection.

Photographs of the species were taken *in situ* using an OLYMPUS LENS camera (24 wide optical zoom ED 4.5-108, 0 mm 1: 3.0-6.9). Their substrate has been recorded. The environmental and macroscopic characterization was based on characteristics such as the color, size and shape of the cap, the outer surface of the cap, the texture of the cap surface, the blades and the presence or absence of latex, the geographical coordinates and that of the host substrate. Macroscopic descriptions were made subjectively and using a description sheet proposed by De Kesel et al. (2002). Other characteristics such as: length and width of the stipe, spacing of the lamellae, presence of volva and ring, etc. were noted. Cécile Lemoine's new mushroom guide; E. Gerhardt's Vigot Mushroom Guide; Eyi Ndong et al. (2011) and a Farvekort brand colour code developed by Peterson (1995), De Kesel et al. (2017) were used for descriptions and identifications. The specimens were dried using a Stöckli DORREX electric dehydrator and stored in transparent Mini grip bags to prevent deterioration under conditions of excessive humidity.

Anatomical studies were carried out on exsiccata (dried samples). Microscopic examination of the tissues was made with 5% KOH and stained with 1% ammoniacal Congo Red. Spores

were observed in the Melzer's reagent to test the amyloidity. This amyloidity is best observed by soaking the tissues overnight in Melzer's reagent and observing in fresh reagent the next morning. Biometric variables such as spores and basidia sizes were measured by using a millimeter ocular lens. At least 50 spores and 20 basidia per sample were measured. Anatomical details were obtained using a NIKON Eclipse Ci optical microscope with a drawing tube, slides and cover plates.

Taxonomy

Amanita citrina (Schaeffer) Persoon

Pileus 40-120 mm in diam., solitary, or rarely in pairs or gregarious; small size with a cap first hemispherical when young, then slightly spread out; it becomes convex when mature and slightly depressed at the center; dry but has a mucilaginous appearance when humid; the flesh is white, full, firm, slightly thick with a smell of fresh flour or tuber and a slightly sweet taste; with a surface almost entirely covered with small lying squamules, adhering from white to yellowish-white, sometimes disappearing when mature and leaving the cap smooth; these squames also form large or small patches. Lamellae, free, 6 to 8 mm high, white, fragile, uneven. Stipe 40-90 mm, cylindrical, central; 5-10 mm thick; bulbous volva in the shape of a circle or even circumcised, sometimes split; well-developed annulus in a superior position, white or yellowish, persistent, skirt shape; white flesh, full.

Spores with two shapes in the preparation, oval and ovoid but generally ellipsoid, 5-8.83-10 × 6-6.42-12 µm ($Q = 1.00-1.40-2$; $n = 30$), amyloid, dense in the preparation. Basidia 23-40 × 9-14 µm, usually small in size, upper portion broad and attenuating downwards to hail, tetrasporic with small sterigmata. Cystidia 26-52 × 9-14 µm, obovoids to uptriforms, rounded apex. Terminal cells 75-47 × 3-9 µm, well differentiated, emerging, long, hairy (Figures 2 and 3). Specimens examined: Burkina Faso. Province of Comoé, Niangoloko, groups at *Isoberlinia doka*, N 10°10'324", W 004°55'752"; alt. 296 m, 20/07/2018, Dabiré K., 305 (holotype).

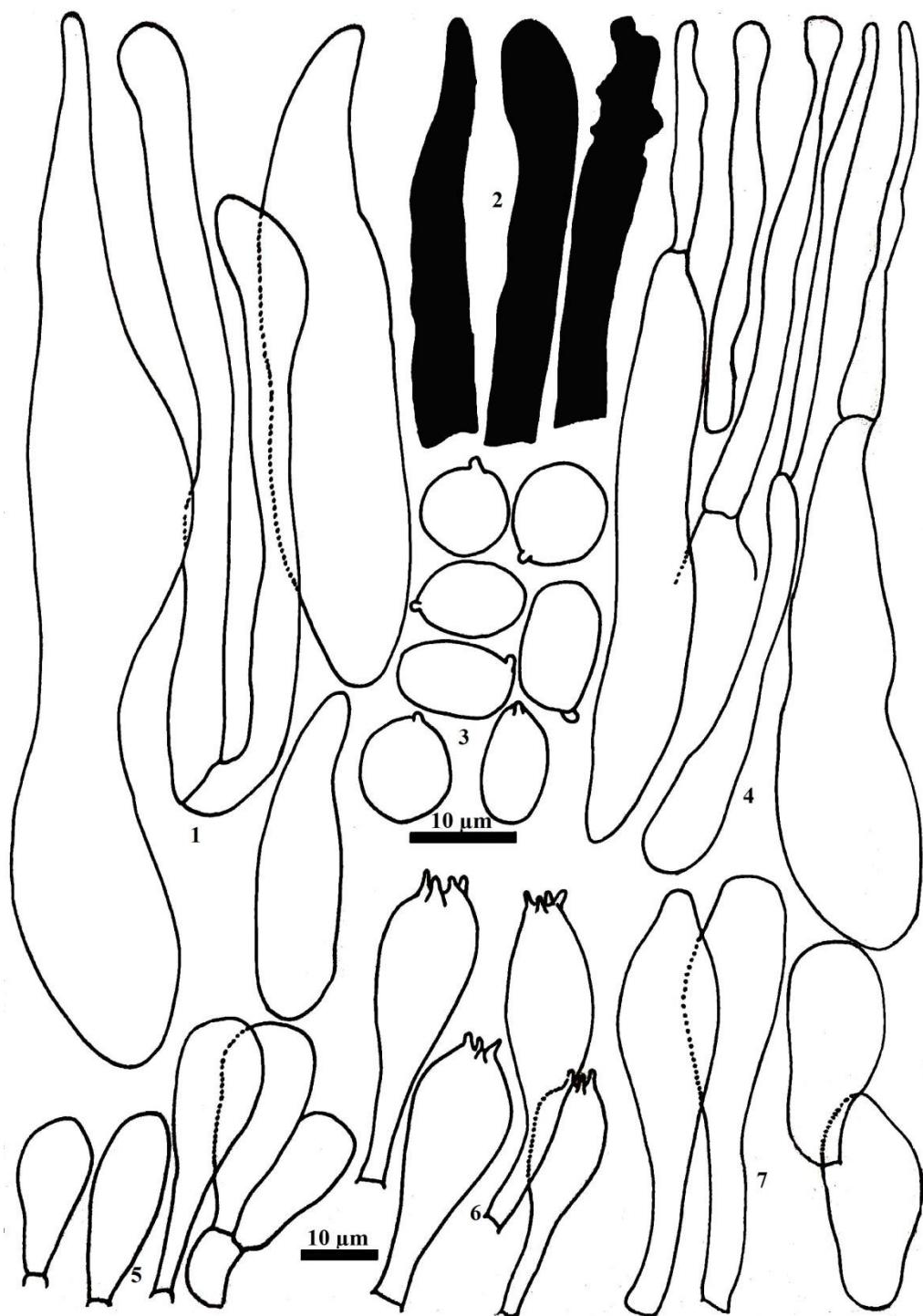


Figure 3. *Amanita citrina*. Dermatocystidia. 2. oleiferous cells. 3. Basidiospores. 4. Terminal cells of the pileus. 5. Basidiola. 6. Basidia. 7. Pleurocystidia. Bar: 10 μm (drawing Dabiré K.)

Amanita pulverulenta Beeli

Pileus 40-180 mm in diam., initially hemispherical, convex to plano-convex; white towards the margin and greyish-white towards the center; surface covered with greyish warts and having a black

colour towards the top; warts fall easily with the slightest movement of wind, rain or animals and leaving the cap smooth; the cap also leaves powdery spots on the cap and even on the hand when touched; white meat. Lamellae, free, white, wide, uneven. Stipe 20-60 mm, subcylindrical, central, powdery, concoloured at the cap,



Figure 4. Basidiomes of *Amanita pulvulenta* photographed in the field.

voluminous when young and thin at maturity; internal structure white-shaded and stuffed; bulbous and rooted volva, sometimes cracked in a few places at maturity; ring well opened out and wide in the shape of a skirt, white, easily falling, from a superior position. Spores elongated to cylindrical, $9-10.42-12 \times 4-5.33-7 \mu\text{m}$ ($Q = 1.5-2.01-2.75$; $n = 30$), plenty, amyloid. Basidia $29-44 \times 7-10 \mu\text{m}$, slightly stalked, not very long, sometimes attenuating towards the base, tetrasporic with sterigmate rather small, rarely 3-sterigmate. Cystidia $26-50 \times 7-10 \mu\text{m}$, rounded apex, upper portion broad or subcylindrical. Terminal cells $2-36 \times 1-6 \mu\text{m}$, not very emergent, quite small, and clamped (Figures 4 and 5). Specimens examined: Burkina Faso. Province of Comoé, Niangoloko, groups at *Isoberlinia doka*, N $10^{\circ}10'406''$, W $004^{\circ}55'827''$; alt. 308 m, 19/07/2018, Dabiré K., 240 (holotype).

Amanita odorata Beeli

Pileus 50-96 mm in diam., mostly isolated and often gregarious (two or three species); convex to plano-convex; greyish-white; small warts strongly welded to the hat, pyramidal with pointed tips, grey; white flesh. Lamellae, white, free, uneven and turning green after harvest. Stipe 70-90 mm, subcylindrical to cylindrical with a thickened base forming a bulb reaching 58 mm high and 55 mm in diameter and rooted; colored at the cap; cracks at maturity at the bulb; fleshy and white flesh; dirty white ring, fragile, membranous and fleeting in a superior position. Spores elongated, $7-8.83-11 \times 3-$

$7.5-7 \mu\text{m}$ ($Q = 1.14-1.94-2.75$; $n = 30$), numerous in the preparation, amyloid. Basidia $22-50 \times 6-10 \mu\text{m}$, hail, tetrasporic with more or less long sterigmata. Cystidia $29-52 \times 7-9 \mu\text{m}$, thin or even hairy, obovate, apex rounded to slightly rostrum. Terminal cells $10-30 \times 2-6 \mu\text{m}$, more or less differentiated, small (Figures 6 and 7). Specimens examined: Burkina Faso. Province of Comoé, Niangoloko, groups at *Isoberlinia doka*, N $10^{\circ}10'324''$, W $004^{\circ}55'752''$; alt. 296 m, 20/07/2018, Dabiré K., 244 (holotype).

DISCUSSION

The biodiversity of mushrooms in Burkina Faso remains poorly known because less attention has been paid to them than to higher plants. The genus *Amanita* Pers. is an example. The three species recorded share the same ecological characteristics, host or substrate. Specifically, most wild mushrooms species belonging to the Russulaceae, Cantharellaceae, Polyporaceae, Boletaceae and Amanitaceae families share the same habitat and ecological characteristics (Chelela et al., 2015); however, *A. citrina* is found in slightly brighter areas while the other two are found in both shadows and light under broadleaved areas. This is due to the characteristics of

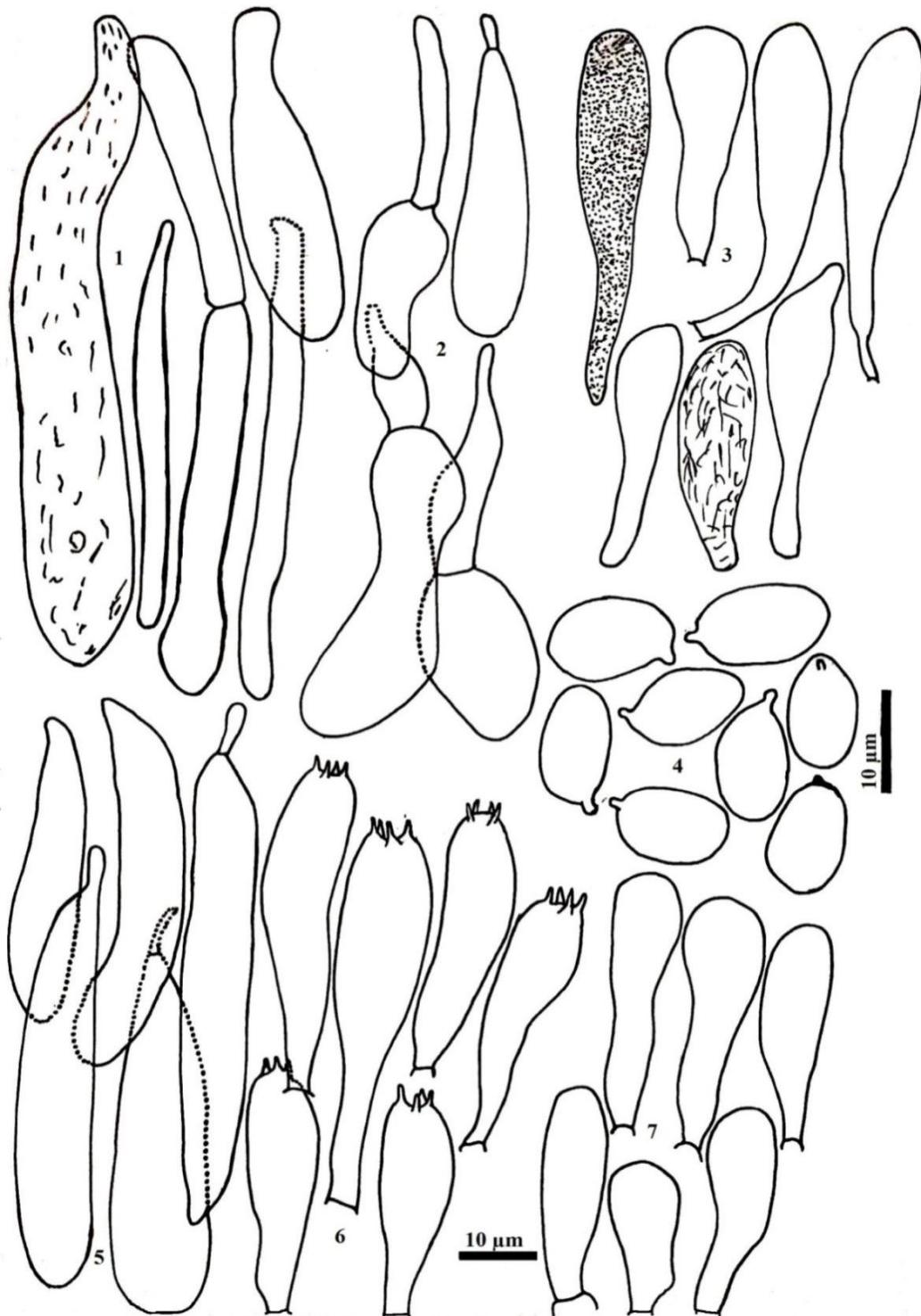


Figure 5. *Amanita pulverulenta*. 1. Dermatocystidia. 2. Terminal cells of the pileus. 3. Pleurocystidia. 4. Basidiospores. 5. Caulocystidia. 6. Basidia. 7. Basidiola. Bar: 10 μ m (drawing Dabiré K.).

the region (Cascades) and the nature of its habitat (Sudanian classified forests with groups of trees of *Berlinia grandiflora*, *Isoberlinia doka*). The high coverage rate of these different plant groups provides shade and

humidity conducive to the growth of fungi.

Amanita pulverulenta shares a morphology almost similar to that of *A. virido-odorata*: macroscopically, the two taxa are characterized by the color and shape of the



Figure 6. Basidiomes of *Amanita virido-odorata* showing cracks in the volva.

warts in the hat and the shape of the stipe; warts in the first species detach very easily leaving the hat smooth and covered with powdery spots while those in the second species are tough, small and fused to the hat. Both this study and other recent studies (Sanon et al., 2017; Dabiré, 2017) exemplify the high diversity of fungal species in Burkina Faso, suggesting that there are still more taxa to be discovered and recorded.

Up to now, most taxonomic research on fungi in Burkina Faso has focused on Russulaceae (Sanon et al., 2014; Sanon, 2015) scleroderms (Sanon et al., 2009; 2011) and in general (Guisse, 2005; Guisse et al., 2015, 2008; Bicaba, 2013) in the western, central plateau, central and central regions and the discoveries of amanite species in the cascade region suggest that further studies are needed in other regions of the country with different mycorrhizal hosts and ecology.

Our own experiences and field observations reveal that amanites have a different phenological appearance. Indeed, they appear well after the other groups of fungi, resulting in a late appearance. This observation was made by Guinberteau and Courtecuisse (1997) where they classified the genus *Amanita* as a late mycorrhizal genus by qualifying them as "early stage fungi". Madamo et al. (2017) add that this phenology would be greatly influenced by humidity. Tulloss et al. (2016) report that *Amanita* is a genus of fungi suspected of being ectomycorrhizal. These three species have been found in some parts of Africa. *Amanita citrina*, *Amanita pulvulenta* and *A. virido-odorata* are known in Benin (Yorou et al., 2017; Yorou et al., 2014; Aignon, 2016; Laourou, 2016), *A. pulvulenta* in Niger (Yorou et al., 2017), *Pulvulenta* in Togo (Kamou et al., 2017; Kamou,

2012; De Kesel and Gueelly, 2007); *A. pulvulenta* and *A. virido-odorata* in Burundi, Congo in Zambia (Bas, 1969; Buyck 1994). Ectomycorrhizal fungi are mainly forest fungi and their best substrates are the presence of ectotrophic species and their roots. These results are similar to those of Zhang et al. (2004) who indicated that the abundance and diversity of ectomycorrhizal fungi are influenced in forest ecosystems dominated by tropical and temperate plants of great economic importance such as Caesalpinoideae, Dipterocarpaceae, and Phyllanthaceae.

Gévry (2010) argues that at the local level, the abundance of precipitation would be an excellent indicator of the diversity and structure of fungal communities. The preservation of the forest influences the subsistence of mushrooms. Fungal distributions are mainly determined by genetic, ecological and morphological constraints (Pringle et al., 2009). For example, ecological and phylogenetic studies have shown that some fungal species may be virtually free of dispersal barriers (Queloz et al., 2011), while others have strong biogeographic structures (Matheny et al., 2009) that may be related to host and habitat limitations.

Conclusion

This study the second of its kind gives an inventory, characterizes and identifies the wild plant genetic resources of fungi of the genus *Amanita* Pers. Mycological investigations have shown that there is a great diversity of *Amanita* species in Burkina Faso, which play a particular role in ecosystems. However, many of

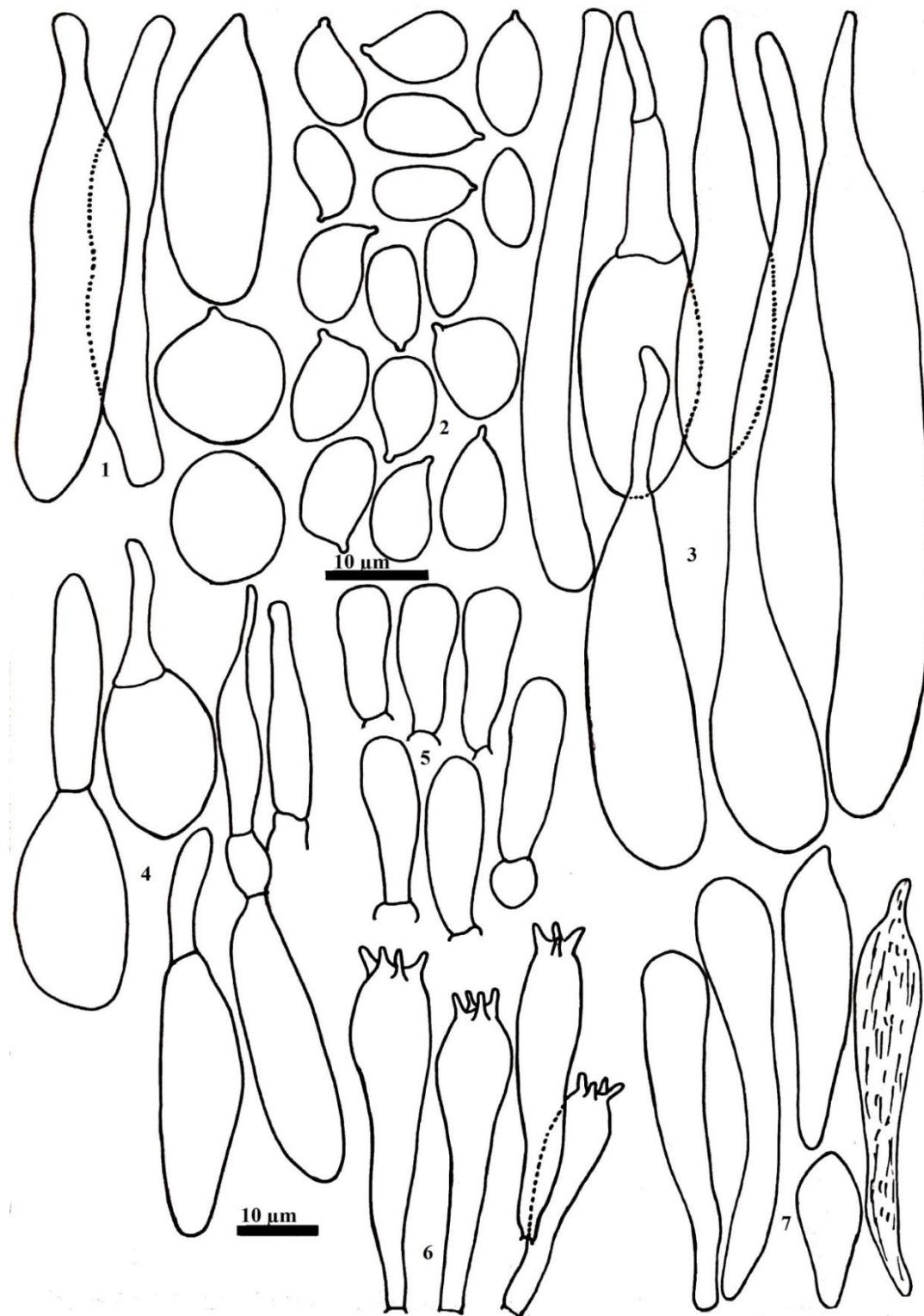


Figure 7. *Amanita virido-odorata*. 1. Dermatocystidia. 2. Basidiospores. 3. Caulocystidia. 4. Terminal cells of the pileus. 5. Basidiola. 6. Basidia. 7. Pleurocystidia. Bar: 10µm (drawing Dabiré K.)

them have not yet been studied and identified. Further research should be carried out to characterize the

biodiversity of Burkina Faso fungi, including Amanitaceae.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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